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Project title: Complete host testing with a potential biological control agent on common reed in view of submitting a petition for field release in winter 2014/15

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14. ABSTRACT <p><b>The perennial grass <i>Phragmites australis</i>, or common reed, is considered highly invasive in North America. The native North American populations of common reed were recently recognized as a distinct subspecies, <i>P. australis americanus</i>. Investigations to evaluate the potential for classical biological control of the invasive reed type started in 1998. The CABI Centre in Switzerland was subcontracted to search for host specific natural enemies in the area of origin of <i>P. australis</i>. During the last few years, work concentrated on the two noctuid moths <i>Archanara geminipuncta</i> and <i>A. neurica</i>. Although both species can develop on native North American reed under no-choice conditions, lower oviposition and lower overwintering survival is expected on native compared to introduced reed. An open-field test established in 2013 with both moths confirmed a strong oviposition preference of <i>A. neurica</i> for introduced reed, but did not yield conclusive results for <i>A. geminipuncta</i>. The current contract provided funding to repeat the open-field test with <i>A. geminipuncta</i> in summer 2014 and to summarize results in preparation of a petition for field release in North America. Females of <i>A. geminipuncta</i> laid 312 (95.7%) of their eggs onto European and introduced reed, and only one egg cluster with 14 eggs on native reed. This clearly confirms the strong oviposition preference of the moth for native reed. Coupled with the fact that the eggs that are laid on native reed suffer a 40% higher overwintering mortality, we believe that any impact of <i>Archanara</i> species on native reed would be negligible. We are currently contributing to the petition for field release of <i>A. geminipuncta</i> and <i>A. neurica</i>, which is being prepared by Richard Casagrande (University of Rhode Island) and Bernd Blossey.</b></p>		
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## Abstract

The perennial grass *Phragmites australis*, or common reed, is considered highly invasive in North America. The native North American populations of common reed were recently recognized as a distinct subspecies, *P. australis americanus*. Investigations to evaluate the potential for classical biological control of the invasive reed type started in 1998. The CABI Centre in Switzerland was subcontracted to search for host specific natural enemies in the area of origin of *P. australis*. During the last few years, work concentrated on the two noctuid moths *Archanara geminipuncta* and *A. neurica*. Although both species can develop on native North American reed under no-choice conditions, lower oviposition and lower overwintering survival is expected on native compared to introduced reed. An open-field test established in 2013 with both moths confirmed a strong oviposition preference of *A. neurica* for introduced reed, but did not yield conclusive results for *A. geminipuncta*. The current contract provided funding to repeat the open-field test with *A. geminipuncta* in summer 2014 and to summarize results in preparation of a petition for field release in North America. Females of *A. geminipuncta* laid 312 (95.7%) of their eggs onto European and introduced reed, and only one egg cluster with 14 eggs on native reed. This clearly confirms the strong oviposition preference of the moth for native reed. Coupled with the fact that the eggs that are laid on native reed suffer a 40% higher overwintering mortality, we believe that any impact of *Archanara* species on native reed would be negligible. We are currently contributing to the petition for field release of *A. geminipuncta* and *A. neurica*, which is being prepared by Richard Casagrande (University of Rhode Island) and Bernd Blossey.

## 1. Introduction

In 1998, a project was started at CABI in Switzerland to evaluate the potential for biological control of common reed in North America. During a two-year survey, 15 sites in Central Europe were sampled for endophagous herbivores of *P. australis*. In a first step, eight moth species and one chloropid fly were prioritized for further investigations as potential biological control agents (Häfliger et al. 2001). Currently, work is focused on the two noctuid moths, *A. geminipuncta* and *A. neurica*.

Common reed, *Phragmites australis* (Cav.) Trin. ex Steudel, is a cosmopolitan, perennial, clonal grass that can form large monospecific stands in wetlands and along rivers and lakesides. Due to its high genetic and morphological variability, *P. australis* is able to grow in a wide range of habitats with different climates (van der Toorn, 1972). In Europe, reed beds are inhabited by a rich insect community and are valuable and endangered ecosystems (Tscharntke, 1999; Tewksbury et al., 2002). In North America and Australia, however, *P. australis* is considered invasive and a threat to biodiversity (Wapshere, 1990; Marks et al., 1994; Tewksbury et al., 2002). Only in the last century did *P. australis* start to spread in North America. Before that, it had been present for at least 3500 years without being invasive. The dramatic increase of common reed populations in the second half of the 20<sup>th</sup> century has often been attributed to land use changes and eutrophication. However, the alternative hypothesis of the introduction of an invasive European genotype was verified by genetic studies of Saltonstall (2002). We now know that there are several native haplotypes in North America, but particularly in the East and Midwest they are usually rare and out-competed by one invasive European haplotype. The native North American populations of common reed were recently recognized as a distinct subspecies, *P. australis americanus* (Saltonstall et al., 2004).

For the development of biological control this means that herbivores are required that are able to reduce the invasiveness of the introduced European type, without adversely affecting the native North American subspecies. Host-specificity tests carried out at CABI and at the University of Rhode Island showed that both *Archana* species have a very narrow host range. Larvae were only able to complete development on plants of *Phragmites* spp. However, larval development tests carried out in 2004 showed similar development rates on native North American and European reed (Häfliger et al. 2005).

Preliminary oviposition tests carried out at CABI in the following years indicated a potential preference of adult moths for European reed. Further experiments were needed to confirm and quantify these findings.

## 2. Open-field oviposition tests with *A. geminipuncta*

### 2.1 Material and Methods

Four plots were set up: 1) 7 pots of native and 7 pots of introduced reed; 2) 14 pots of introduced reed; 3) 14 pots of native reed; and 4) 14 pots of European reed (see Fig. 1). Between five to eight different populations of reed per origin were randomly exposed. Each plot was placed in the corner of a 10x10 m quadrat and 11 females of *A. geminipuncta* were released per plot, i.e. 44 females in total. Two weeks after the last release, all stems were harvested and checked for eggs.

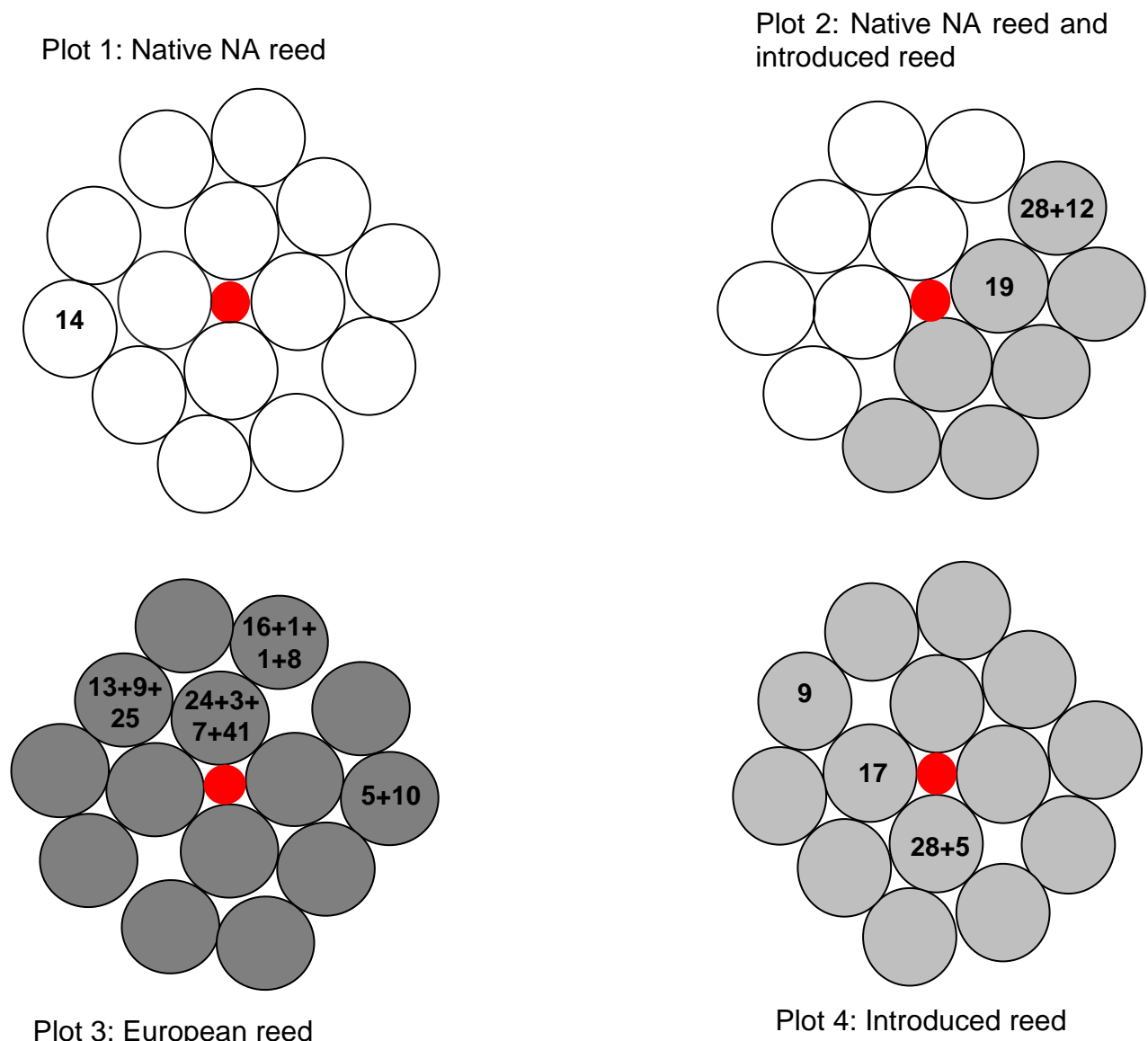
**Table 1** Origin of plants used in the open-field oviposition test with *Archanara geminipuncta* in 2014.

Population	Origin	Number of pots 2014
Astoria, OR	Native	2
Beldens Landing, CA	Native	2
Montezuma, NY	Native	3
Saratoga Springs, UT	Native	5
Savage Fen, MN	Native	4
Seminary Fen, MN	Native	1
Spring Bluff, IL	Native	1
Sun Lake Park, WA	Native	3
Assunpink Lake, NJ	Introduced	3
Cape May, NJ	Introduced	4
New Haven, CT	Introduced	5
Novato, CA	Introduced	4
Rock Ford, WA	Introduced	1
Saratoga Springs, UT	Introduced	2
Delémont, Switzerland	European	4
Magadino, Switzerland	European	2
Yverdon, Switzerland	European	2
Hodmezovasarhely, Hungary	European	3
Iasi, Romania	European	3

### 2.2 Results

We found 326 *A. geminipuncta* eggs on the exposed shoots. *Archanara geminipuncta* laid 95.7% of its eggs onto European and introduced reed. Only one egg cluster with 14 eggs was found on native reed, despite the fact that native reed shoots had larger stem base diameters than European and introduced reed (Table 2).

**Figure 1** Setup and results of open-field oviposition test with *A. geminipuncta* in 2014 (red dot = Moth release points; numbers indicate number of eggs laid on individual shoots).



**Table 3** Results of open-field oviposition test with *Archanaara geminipuncta* in 2014.

	Mean			# egg batches /
	# stems per pot	stem length (cm)	stem diameter (mm)	# eggs
European	22.0	116.7	3.4	16/194
Introduced	18.2	98.4	3.5	7/118
Native	15.4	122.7	4.1	1/14*

\* we found 2 additional egg batches with a total of 39 eggs. However, we excluded these eggs, because they were laid on shoots that showed obvious characteristics of European/introduced reed and must have been entered the pot via rhizomes that had grown in from neighboring plants.



### 2.3 Discussion and Conclusions

Previous no-choice larval development and oviposition tests with *A. geminipuncta* showed only minor differences in acceptance between the native and the introduced subspecies of *P. australis*. However, the data presented here clearly show that both moth species strongly prefer invasive reed over native reed for oviposition when given the choice in a semi-natural field setting. Leaf sheaths of native reed are looser and tend to fall off before winter. Ovipositing females might notice the difference and avoid these stems.

In addition, our overwintering experiment carried out in 2006/2007 suggests that any eggs laid on the native subspecies *P. australis americanus* would suffer higher mortality due to differences in phenology of the two *P. australis* subspecies. Eggs laid on *P. australis americanus* will mostly fall off together with leaf sheaths before or during winter and be exposed to climatic conditions, predators, and pathogens. The overwintering experiment was carried out in a common garden. We expect mortality of unprotected eggs to be even higher under field conditions. Especially on water reed, eggs might experience increased mortality due to flooding, eggs might get washed away, or changing moisture regimes may increase fungal attack. Together with the low number of eggs found on native reed, we expect any impact of *A. neurica* and *A. geminipuncta* on native reed to be negligible, should the noctuids be released in North America. We are currently contributing to the petition for field release of *A. geminipuncta* and *A. neurica*, which is being prepared by Richard Casagrande (University of Rhode Island) and Bernd Blossey.

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